PGPR: A POTENTIAL ALTERNATIVE TO METHYL BROMIDE FUMIGATION IN VEGETABLE PRODUCTION

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Methyl bromide fumigation applied before planting is a widely used weapon for disease, nematode and weed control in U.S. vegetable production. Because the 1990 Clean Air Act has decreed that methyl bromide use will be phased out by the year 2001, alternative pest control methods are desperately needed that meet the requirements imposed by high value vegetable production. One alternative for plant disease management is the use of biological control agents. Plant growth-promoting rhizobacteria (PGPR) represent one group of biological control agents which can be practically used in agriculture because they can be delivered by seed treatment, transplant drench application, or soil mix inoculation.

Research over the past two decades has demonstrated that plants have latent defense mechanisms against pathogens which can be systemically activated by exposure of plants to stress or infection by pathogens. This phenomenon, called systemic acquired resistance or induced systemic resistance, operates through the activation of defense genes and the accumulation of defense compounds at a site distant from the point of pathogen attack. We previously demonstrated that treatment of seeds or roots of cucumber with select strains of PGPR induces systemic resistance to multiple diseases of cucumber, and surprisingly, to the cucumber beetle vector of bacterial wilt disease. However, these studies were done in fumigated soil, and the effects of soil sterilization on PGPR-induced resistance are not known. The objectives of this study were to evaluate PGPR-induced disease resistance in cucumber with and without methyl bromide fumigation, to evaluate multiple PGPR applications for growth promotion effects and protection against bacterial wilt disease in cucumber, and to determine if PGPR could be used as an alternative to methyl bromide fumigation to promote early growth which may be inhibited by soil borne disease pathogens.

Field experiments were conducted in 1994 and 1995 at the E.V. Smith Horticulture Substation in Shorter, AL. In: 1994, studies were done to compare PGPR treatment, with and without methyl bromide fumigation, to weekly applications of insecticide (esfenvalerate) for control of bacterial wilt disease of cucurbits caused by *Erwinia tracheiphila*, a pathogen vectored by cucumber beetles. Plots consisted of two, 9 meter-long rows of cucumber seeded on 12 April. Treatments were replicated 4 times in a randomized complete block design and included 3 levels of bacterial wilt control (PGPI, insecticide and nontreated control) with or without fumigation (393 kg/ha of 67% methyl bromide + 33% chloropicrin). The fumigant was injected into raised beds followed immediately by application of black plastic mulch. 'Straight 8' cucumber seeds were dipped into pelleted bacterial cells (PGPR treatment) or into distilled water (nontreated control) before planting. The numbers of cucumber beetles and wilted vines per plant in

the treatment plots were recorded weekly, and the total harvested fruit weight per plot was determined. The 1995 experiment was done to evaluate PGPR treatments, with and without fumigation, for stimulation of early plant growth, which is inversely related to infection by *Pythium* and *Rhizoctonia* damping off diseases, and for protection against bacterial wilt disease. Plots consisted of one, 9 meter-long row of 'Straight 8' cucumber planted on 27 April. Treatments were replicated 6 times in a randomized complete block design and included 5 levels of PGPR treatment (PGPR soil drench at planting plus additional PGPR soil applications at 2, 3 and 4 weeks after planting and a nontreated control), with and without fumigation as described above. Plant height was measured 21 days after planting and the incidence of bacterial wilt symptoms was recorded weekly.

In 1994, PGPR treatment was as effective as weekly insecticide applications for control of cucumber beetles (Table 1). We have previously shown that a possible mechanism for the negative effects of PGPR treatment on cucumber beetle feeding is a reduction or decreased mobilization of cucurbitacin, a tri-terpenoid compound found in cucurbits that is a strong cucumber beetle feeding stimulant. PGPR-induced resistance occurred in both fumigated and nonfumigated soils. However, greater disease protection occurred in nonfumigated soils (10. 8% wilted vines in the PGPR plots without fumigation, compared with 27.7% in PGPR plots with fumigation), indicating that fumigation has a negative effect on PGPR-induced resistance to bacterial wilt. Cucumber yields were highest in the PGPR-nonfumigated treatment.

The 1995 results indicated that the at-planting and "booster" applications of PGPR resulted in increased plant growth in both fumigated and nonfumigated plots, compared with the nonbacterized control (Table 2). However, in nonfumigated soils, multiple PGPR applications resulted in greater disease protection than the single, at-planting application. The classically reported poorer early plant growth in nonfumigated plots, compared to fumigated, was overcome by PGPR. Plant growth in the nonfumigated PGPR treatments was equivalent to plant height measurements in the fumigated, nonbacterized treatments; evidence that PGPR is an effective alternative to methyl bromide fumigation for early plant protection against damping off disease. As in 1994, the percentage of wilted vines was lower in the PGPF, nonfumigated plots than in the PGPR, fumigated plots (PGPR x fumigant interaction significant at P=0.06) (Table 2). Therefore, PGPR-induced protection against bacterial wilt appears to be inhibited by fumigation, while growth promotion by PGPR is enhanced by fumigation.

In summary, these results demonstrate that PGPR induced resistance against bacterial wilt disease occurred in both fumigated and nonfumigated soils, although greater disease protection occurred in nonfumigated. soils. This suggests that soil sterilization has a negative effect on PGPR, possibly by elimination of symbiotic soil microfauna. In addition, the results demonstrated that PGPR treatment compensated for poor early plant growth in nonfumigated soil, evidence that PGPR may be an effective alternative to methyl bromide fumigation in cucumber production for control of damping off disease. Although it is not likely that PGPR could be used as a single replacement for methyl bromide fumigation, PGPR can be used as an IPM component in vegetable production to protect against pests and disease.

Table 1. Effect of PGPR and insecticide treatment with and without MeBr fumigation on incidence of cucumber beetles and bacterial wilt infection, and yield of cucumber, 1994.

oucterial with infection, and yield		No. beetles/plant % Wilted vines		ted vines	Fruit wt. (kg/plot)			
Treatment	MeBr	NF	MeBr	NF	MeBr	NF		
PGPR strain	1.0	0.8	27.7	10.8	2.9	3.3		
90-166								
Insecticide	1.0	1.0	27.3	31.3	3.0	2.8		
(esfenvalerate)								
Nontreated	2.6	2.0	43.0	36.7	2.2	2.4		
control								
Contrast Analysis								
Variable comparison	F	P	F	P	F	<u>P</u>		
MeBr vs no MeBr	3.72	0.054	2.31	0.12	0.18	0.67		
(all treatments)								
MeBr vs no MeBr	NA	NA	5.67	0.02	1.07	0.29		
(PGPR treatment)								
PGPR vs control	82.05	0.0001	14.34	0.0002	8.91	0.003		
Insecticide vs control	75.19	0.0001	3.72	0.055	4.69	0.03		
PGPR vs insecticide	0.15	0.70	3.68	0.056	0.67	0.41		

MeBr=fumigated with MeBr; NF=not fumigated

Values in the upper table are season averages. Beetles were sampled from 20 plants per treatment on 6 sample dates. Bacterial wilt incidence was determined by recording the % of wilted vines on 64 plants in each treatment on a single sample date just before harvest. Fruit yield was determined by weighing all marketable fruit in each plot (4 plots per treatment) on 10 sample dates.

F and P values in the contrast analysis table refer to F and P statistics

Table 2. Effect of multiple PGPR applications with and without MeBr fumigation on plant growth and incidence of bacterial wilt, 1995.

	Average p	% Wilted vines/plant (AUDPC values)		
PGPR treatment	MeBr	NF	MeBr	NF
1. Soil drench at planting	13.4	10.0	9.5	8.9
2. (1) + soil drench I wk after	12.6	9.6	11.1	8.7
planting				
3. (2) + soil drench 2 wk after	15.0	10.1	12.1	5.5
planting				
4. (3) + soil drench 3 wk after	14.2	10.5	11.6	6.2
planting				
5. Nontreated control	10.1	6.9	15.3	10.3
	Factorial Analysis	of Variance		
Effect	F	P	F	<u>P</u>
PGPR treatment	72.42	0.0001	5.75	0.0001
Fumigation	467.94	0.0001	19.01	0.0001
PGPR x fumigation interaction	2.85	0.015	2.13	0.06

MeBr=fumigated with MeBr; NF=not fumigated Plant height was measured 21 days after planting

Mean % wilted vines per plant recorded on 5 sample dates (50, 57, 64, 71, and 78 days after planting) on 16 plants per plot (64 plants per treatment) and area under disease progress curve (AUDPC values were calculated to determine bacterial wilt disease progression.

F and P values in the analysis of variance table refer to F and P statistics